VI Spirochaetal Jaundice
Appendix No. 2.

951

WEIL'S DISEASE (SPIROCHÆTOSIS ICTERO-HÆMORRHAGICA) IN THE BRITISH ARMY IN FLANDERS

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A SHORT preliminary note[1] on infectious jaundice among British troops in France was written in the early summer of 1916. Although the investigation of this disease is still being pursued both here and elsewhere and is as yet far from complete, the accumulation of considerably more material, both clinical and experimental, since our last publication would seem to justify a further communication at the present moment. The additional results which we have attained contain little not already described by Inada and his collaborators, but they seem of interest in that our confirmation of the findings of the Japanese workers, at the time of our first note only partial, has now been rendered almost complete. The only results which we have not been able to duplicate are their success in growing the causal organism in artificial culture and the finding of spirochætæ in the urine. The possible explanation of the former point and the evident explanation of the latter are discussed below.

Previous Work.

The disease commonly bears the name of Weil, who described it in 1886, but Martin and Pettit, in a recent publication discussed below, point out that this is quite unjust in that several French authors, quoted from Kelsch (1894), had previously described the condition under various names, and at dates extending from the siege of Cairo down to 1886. Those quoted are: Larrey, Ozanam (1849), Monneret (1859), Laveran the elder (1865), Lancereaux (1882), Landouzy (1882), and Mathieu (1886).

* Received for publication on Dec. 8th, 1916.

[1] Journal of the R.A.M.C., September, 1916, and Brit. Med. Jour., Sept. 23rd, 1916.

Weil described the chief characteristics of the disease as being jaundice, pyrexia, and hæmorrhages, and the occurrence of cases in epidemics or localised groups. Widespread epidemics have been recorded in the United States, and also outbreaks in India, Africa, and the Near East. In Osler's Medicine we find :—

The symptoms are at first gastric, then fever follows (with the usual concomitants) and jaundice, which may be slight or very intense, and, as a rule, albuminuria. The liver and spleen are often enlarged, and in the severe forms there are nervous symptoms and hæmorrhages. There is often a secondary fever, and the atacks last from ten days to three weeks. The course is usually favourable ; fatal cases are rare in the United States and in India and South Africa, but in the Greek hospital at Alexandria the death-rate was 32 among 300.

No definite etiology had been established until the subject of infectious jaundice was reopened by a monograph from Drs. Inada, Ido. Oki, Kaneko, and Ito, which was first published in the English language in March, 1916, in the *Journal of Experimental Medicine*. In this paper the authors state :—

In the western part of Japan there prevails an epidemic and endemic disease, characterised by conjunctival congestion, muscular pains, fever, jaundice, hæmorrhagic diathesis and albuminuria, which is known as Weil's disease or febrile jaundice. At the end of last year (1914) the same disease was observed in Shiba, in the eastern part of Japan near Tokio, where the patients numbered 178.

In 1914 Inada and Ito reported the discovery of a spirochæta in the liver of a guinea-pig which had been injected with the blood of a patient suffering from Weil's disease. They reported that they were able to repeat this finding with the blood of other cases, the animals developing albuminuria, pyrexia, and jaundice, and they were able to show the spirochætæ in the liver and blood of the animals in large numbers. They were able to pass the infection from animal to animal, and in one strain they had reached 50 passages. The experiments made with the blood of normal people and of patients with catarrhal jaundice produced no such result. Subsequently the Japanese workers were able to demonstrate "the same spirochætæ in six specimens of patients' blood, in the intestinal wall of one case, and in the adrenal gland of another case, of 11 autopsied cases." In two cases which came to autopsy on the sixth day of illness they found the spirochætæ in the liver in almost as great numbers as in the liver of guinea-pigs. Finally they showed that the blood serum of patients recovering from the disease contained protective substances against the spirochætæ.

From these data they came to the justifiable conclusion that the spirochæta which they had found was to be considered the causal organism of the disease. They state that they have, with the blood of 13 out of a series of 17 cases, succeeded in producing the disease in guinea-pigs, the animal showing the typical signs of the disease and the spirochætæ being found in the blood and liver. The blood from all cases in which it was taken on the fourth or fifth day of illness gave positive results, but after the fifth day the results became more inconstant as the disease progressed. They indicate that the spirochætæ disappear from the blood coincidently with the appearance of immune substances in the serum. In the early stages the spirochætæ were in the blood stream, but as the disease progressed they disappeared from the blood, and the authors were not able to demonstrate them in the tissues. At some period after the tenth day spirochætæ appear in the urine and may be demonstrated both by dark-ground illumination and by animal inoculation. They consider the appearance of the spirochætæ in the urine to depend on the development of an immune substance in the serum. They have been able to find spirochætæ in the urine as late as the thirtieth day.

Weil believed that the entry of the infection was through the alimentary canal, and Inada supports this view. Ido and Oki have noticed that the disease sometimes begins with a local swelling of the lymph glands, and, further, they have been able to communicate the disease to animals by applying infected material to the uninjured skin. This led them to think that there was a possibility of infection occurring through the skin. In support of this view they note that the disease was more frequent in the wet parts of certain mines, and that, the water having been pumped out on their suggestion, no further cases occurred in those particular areas. They also noted that there were more cases in wet mines than in dry, and that men working on the surface did not contract the disease. They think from clinical observation that direct infection from man to man is rare. Without stating their reasons they say that they consider transmission by mosquitoes or by vermin unlikely.

In the *Bulletin de l'Académie de Médecine*, Oct. 10th, 1916, Dr. L. Martin and Dr. A. Pettit have published an interesting note confirming the existence of the disease in the French Army. They have followed two cases in full and succeeded in finding the spirochætæ in the urine and in infecting with the urine of patients convalescent from the disease. They further add an important link to the epidemiology of the disease in describing a most interesting case of one of their colleagues, who himself became infected in passing the disease from animal to animal, the circumstances leaving no doubt that the period of incubation was six to eight days.

General Clinical Picture of the Disease.

Our attention was first called to the disease by a fatal case of jaundice under the care of Captain Flood, R.A.M.C., whose permission we have for mentioning the case.

The man was admitted with a temperature of 105° F. and very deep jaundice. On the evening of admission there was severe epistaxis, which necessitated the plugging of the nares. Hæmorrhagic diarrhœa set in, and on the third day after admission the trunk and limbs were covered with petechiæ. Death followed on the next day. Multiple hæmorrhages in the pleuræ, pericardium, and peritoneum were the most striking feature, apart from the extreme jaundice of all the viscera. The bile passages were free and the duodenum normal. This is the only case we have seen in which the generalised hæmorrhages occurred in more than trivial degree, and in this case hæmorrhagic diarrhœa was apparently the immediate cause of death. At the time there were no guinea-pigs available for inoculation and the tissues from the case were unfortunately lost. An emulsion of the liver and kidney examined by dark-ground illumination revealed no spirochætæ.

A second case, also fatal, occurred a few days later; the patient was admitted moribund under the care of Major Young, C.A.M.C., and died within a few hours. The autopsy showed nothing significant with the exception of the extreme jaundice of the viscera and advanced hæmorrhagic nephritis. Tissues examined for us by Dr. Laidlaw at Guy's Hospital did not reveal any spirochætæ, and this is in accordance with the findings of the Japanese workers who state that it is very difficult to find the spirochætæ when the patient dies late in the disease, as happened in this case which terminated on the twelfth day.

Since then about 100 cases have been observed by us, and of these 50 have been under the personal care of one of us (J. A. R.). In 26 cases we have infected animals from the patients. The animals have died or have been killed, and have all shown the characteristic picture of the disease in the animal, and we have been able to find the spirochætæ in all of them, either in films or sections of the tissues. Early in our observations we were struck by the occurrence of cases showing a similar clinical picture, but no jaundice, and we have since been able to establish the fact that cases of spirochætosis occur without external signs of jaundice, which would seem to cast a doubt on the accuracy of the terms epidemic jaundice and spirochætosis ictero-hæmorrhagica. We cannot say definitely in how many cases of Weil's disease jaundice occurs as a clinical sign, but we estimate that it does not occur in more than 60 per cent. of cases in the epidemic with which we are dealing.

The onset.—The history of onset was strikingly uniform, and the acuteness of the inception of the disease may be judged from the fact that the patients almost invariably

were able to date the onset by some simple reference to the duty they were performing at the time. The onset is very commonly heralded by a shivering fit and a feeling of faintness or giddiness. Characteristic early symptoms have been vomiting, headache, and pains in the back and limbs. The patients complain of great weakness and of feeling as if they had been beaten all over, and they are so sensitive to these pains that they are reluctant to move their limbs or to turn over in bed. Pain in the eyes is a frequent early symptom, and there are often vague unlocalised abdominal pains. The onset is generally with fever of a more or less high degree, varying between 103° and 105° F. The limb pains appear to be muscular, but in one case were especially referred to the tibiæ. All the patients on admission were very weak and often collapsed, and complained of the early feeling of unsteadiness and inability to stand.

Alimentary system.—The tongue is excessively dry, brown in colour, frequently fissured, and there are sordes on the lips. Herpes labialis, invariably becoming hæmorrhagic, occurred in 42 per cent. of cases. Vomiting in the early stages may be said to be a constant symptom; it often persists for several days, and the vomitus is frequently bilious. A history of blood in the vomitus is occasionally elicited, and streaks of blood have been seen, but as this may have been swallowed from the naso-pharynx it was not noted as a true hæmatemesis. Appetite, which was lost during the pyrexial period, returned as the jaundice faded. Hiccough occurred in the two most severe cases, one of which terminated fatally.

Except in two cases, in which there was a history of initial diarrhœa, extreme constipation was the rule throughout the acute stage of the disease. The stools were usually bile-stained. One fatal case succumbed to a terminal hæmorrhagic diarrhœa, and in one other small traces of blood were observed in the stools. Intestinal hæmorrhages may therefore be regarded as exceptional. During the first few days many patients complained of vague abdominal discomfort and pain, but there was never any localised tenderness. Abdominal examination revealed distinct hepatic enlargement in one case without jaundice, but in no other case was the liver palpable. The spleen was not palpable in any case while under observation here.

Circulatory system.—The pulse-rate is usually slow in proportion to the temperature, and this holds good even in the cases without jaundice. In one severe case the pulse-rate rose to 120 during the first week. In early convalescence there is a definite diminution in the pulse-rate. Even in the bad cases the tension and volume are good. Marked dicrotism was once noted. Except in one case in which the apex beat was for a short time external to the nipple line, there was no clinical evidence of cardiac dilatation.

Respiratory system.—There is, as a rule, no evidence of respiratory complications beyond slight bronchitis in the severer cases or hæmoptysis. The respiratory rate is sometimes raised during the early stages, but there was never any evidence of consolidation or pleurisy. In 26 per cent. of cases there was blood-tinged expectoration. Both "bright-

red" and "rusty" sputa were noted, the latter being the more common. In the two cases in which hiccough was noted a peculiar catchy type of inspiration occurred, and in two others there was an expiratory grunt, although no other pneumonic symptoms were present. Epistaxis occurred in 17 per cent. of cases.

Excretory system.—Two cases had difficulty in micturition for which there was no obvious cause. The urine of jaundiced cases contains bile in varying quantities. In some the urine is very deeply coloured, in others the colouration is less evident. In some of the cases without jaundice bile has been demonstrable in the urine. Albuminuria was almost a constant accompaniment of the pyrexial period and varied from a "faint trace" to a "considerable amount." Granular tube casts and red blood cells were present in centrifugalised specimens. Probably as a result of the fever and vomiting acetone was also demonstrable in the urine of all the graver cases.

Nervous system.—Weakness and great prostration are characteristic of the onset of the disease. Headache and generalised pains and stiffness are constant symptoms, and the pains in the back and legs are exceptionally severe, frequently causing sleeplessness. In the graver cases torpor was noticed, and in one fatal case this progressed to the "typhoid state," and was accompanied by rambling delirium, tremors, and muscular twitchings. Pruritus, probably due to the jaundice, occurred in three cases. The eyes are painful and show very marked injection of the conjunctivæ, and in one case during convalescence there was a pink suffused zone round the sclero-corneal junction, associated with photophobia, and suggesting a definite scleritis. Subconjunctival hæmorrhage occurred in one case.

Lymphatic system.—There is a slight enlargement of the glands. The pectoral group of the axillary glands are sometimes felt as discrete shotty nodules on the thoracic wall of the axilla. The cervical glands are also enlarged and sometimes tender. The spleen has never been palpable.

The skin.—Varying degrees of jaundice are seen. The colour is usually of a lemon-yellow to a deep orange tint, and never approaches the greenish colour of obstructive jaundice. It first appears on the fourth or fifth day of illness, occasionally earlier: it becomes progressively deeper until the ninth or tenth day, and then, as a rule, fades very rapidly. In some cases, however, the colouration is more persistent. Of 47 cases diagnosed as Weil's disease clinically, or by communication of the disease to the animal, 74 per cent. have developed jaundice, and 26 per cent. have not shown any colouration.

Hæmorrhagic herpes labialis, occurring in 45 per cent. of cases, was frequently of very wide extent, in some cases covering the chin and upper part of the neck. In one case the herpes formed a black, confluent eruption which completely surrounded the mouth and invaded the nares; the contrast of the black sores with the brilliant orange colour of the facies, together with the blood-shot eyes, gave the patient an alarmingly morbid appearance. Petechial

hæmorrhages, widely distributed over the trunk, were present in association with the jaundice in one of our cases which recovered, and in one fatal case under Captain Flood. Profuse urticaria occurred in one of our cases.

The fever.—There is an irregular pyrexia descending by lysis, and ranging for the first week between 100° and 103°. Temperatures of 104° and over are common at the onset. The fever terminates between the tenth and fourteenth days, and is often succeeded by a subnormal temperature for three or four days. In four cases there was a relapse of the pyrexia but not of the other symptoms, but it must be remembered that the cases are evacuated at an early date, as soon as the pyrexial period is passed, and it may be that relapses occur more commonly than would appear from our observations. We insert some typical charts (Nos. I.–IX.) of jaundiced and non-jaundiced cases.

Differential diagnosis.—There is seldom any difficulty in distinguishing between Weil's disease and other forms of jaundice. The high pyrexia, prostration, and conjunctival congestion, on the one hand, and the absence of any localising symptoms, on the other hand, exclude catarrhal jaundice, gall-stones, and cholecystitis. Jaundice due to typhoid or paratyphoid infections is to some extent excluded by blood culture, which is usually done on admission. In the early stages, however, before the appearance of the jaundice and in the non-icteric cases the diagnosis is more difficult. The combination of pyrexia, herpes, and hæmoptysis has not infrequently resulted in the patient being sent to hospital with the diagnosis of pneumonia, while others have been labelled pulmonary tuberculosis, suspected cerebro-spinal meningitis, and "trench fever." Between severe forms of "trench fever" and non-icteric spirochætosis it is almost impossible to distinguish during the first few days unless the presence of vomiting and albuminuria in the latter and their absence in the former should prove constant.

Prognosis.—During the first two months in which we had cases under observation there were two deaths, and though during the following three months four-fifths of the total cases have passed through there have been no further deaths previous to evacuation as convalescent. We have heard of two cases which were evacuated during the pyrexial period and have succumbed elsewhere. On the whole, the prognosis may be said to be favourable even in the more severe cases. The death-rate is, so far as we know, less than 6 per cent.

Treatment.—The treatment at present is purely symptomatic. Good nursing, unlimited fluids, alkaline draughts, fruit to eat, and the rectal administration of glucose (6 per cent. solution, 1 pint) once or twice daily to cases with

CHART I.

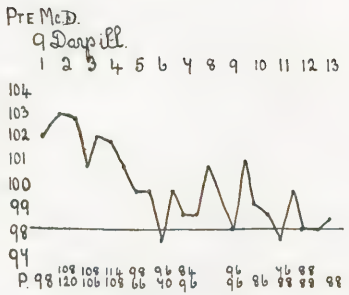


CHART II.

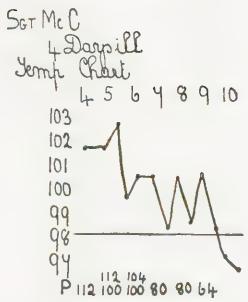


CHART III.

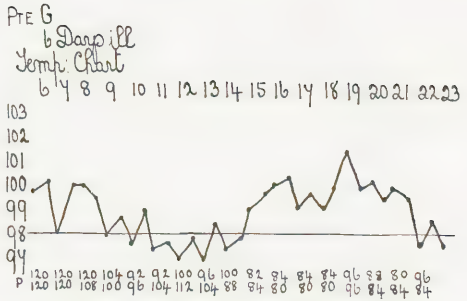


CHART I.—Private McD. Complete chart from first day. History of sudden onset, with shivering, severe pains in head and limbs, jaundice, hæmorrhagic herpes, dry, brown tongue; no enlargement of spleen or liver.

CHART II.—Sergeant McC. Very extensive hæmorrhagic herpes. Jaundice appeared on fifth day. Diagnosed clinically before jaundice appeared. Temperature chart of animal infected from this patient is given later. (See Chart XIII.)

CHART III.—Private G. Chart showing relapse. Vomiting, jaundice, epistaxis, blood-stained sputum, and enlarged glands; hiccough severe, drowsiness and torpor. On the tenth day jaundice began to fade. No return of symptoms, with exception of malaise, with recrudescence of pyrexia.

CHART IV.

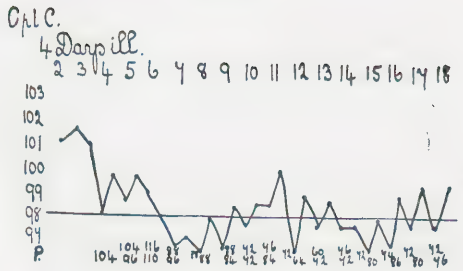


CHART V.

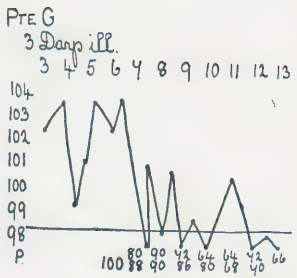


CHART VI.

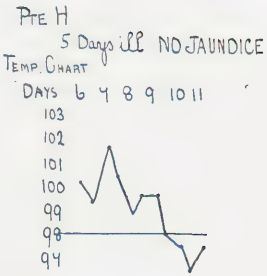


CHART IV.—Corporal C. Chart showing short relapse. Jaundice, herpes, vomiting, epistaxis, albuminuria, acetonuria.

CHART V.—Private G. No jaundice; clinically diagnosed; intense headache, pains in legs and abdomen; liver slightly enlarged and tender; prostration great, injected eyes, albuminuria.

CHART VI.—Private H. Pains in back and legs, injected eyes, drowsiness; no jaundice. Temperature chart of animal infected from this patient is given in the section on the disease in animals. (See Chart XII.)

CHART VII. — Corporal McG. Non-jaundiced case. Admitted second day. Severe pains in head, eyes, back, limbs; albuminuria, hæmoptysis, vomiting. Chart of animal infected from this patient given later. (See Chart XI.)

CHART VIII.—Private H. Fatal case. Aged 40. Admitted on tenth day with deep jaundice, general malaise, headache, and pains in the legs; onset with vomiting. On admission, conjunctivæ injected, tongue dry and furred; liver and spleen not felt; albuminuria. Twelfth day: Mental confusion, retention of urine, hiccough. Thirteenth day: Rambling delirium, retching, jaundice deeper, pruritus; dyspnœic attack. Fourteenth day; Incontinence of urine and fæces, muscular twitchings. Death supervened.

CHART IX.—Private E. Fatal case. Chart of animal infected from this case is given later. Admitted with jaundice and very ill; unable to give clear account of duration of illness; tongue dry and furred, eyes injected, generalised muscular pains; no enlargement of liver or spleen. Patient given alkaline drinks and 1 pint of 6 per cent. glucose per rectum. Next day patient worse; tremors, restlessness, retention of urine, and feeble, intermittent pulse; 1½ pints of 2 per cent. sodii bicarb. intravenously. Died two hours later, probably on the eighth or ninth day of illness. (See Chart X.)

CHART VII.

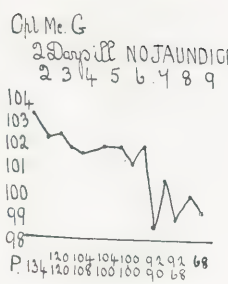


CHART VIII.

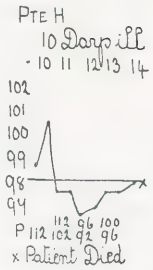
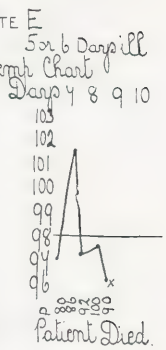


CHART IX.



vomiting and acetonuria constitute the main elements of treatment in our series. When possible the patients are nursed in the open air with apparent benefit.

Clinical Pathology.

In order to attempt to ascertain if the jaundice was of hæmolytic origin we made daily blood counts in four early cases, the count being done on five successive days from the fifth to the ninth day of illness. We were unable to demonstrate any progressive diminution in the cell count or in the hæmoglobin, and the average of the counts was high. Leucocyte counts in the early cases ranged from 9000 to 10,000 cells per c.mm. Stained films did not show any degeneration forms or polychromatophilia, changes which are easily demonstrated in the infected animal. The coagulation-time was taken by Wright's method and ranged around 3 minutes.

The resistance of the red cells to various concentrations of salt solution was tested, and it was found that in the cases with jaundice the resistance was increased. Hæmolysis began in normal blood in 0·450 per cent. NaCl, and did not commence in patients with jaundice till 0·350 per cent., and was not complete at 0·300 per cent. This was not found to be the case in patients suffering from Weil's disease without jaundice, and by the addition of small traces of bile to normal blood cells it was possible to increase their resistance. The end-point of the hæmolysis was very low, "complete" being reached in 4 cases only at 0·250 per cent., whereas the normal controls were "complete" at 0·300 per cent. Blood cultures for enteric group organisms were made in over 50 cases, and were consistently negative at the end of three days.

We searched diligently in blood films for the spirochætæ, using different methods and staining in different ways. In one case after citrating a large quantity of blood (20 c.c.), and allowing the cells to settle, and then centrifuging the citrated plasma, we found one spirochæta and in confirmation the animal infected with the blood developed jaundice. This blood was taken on the fifth day of the disease. Dr. A. C. Coles examined films from two cases for us by his special method; in one, the films being made on the second day of illness, he was able to find two spirochætæ in six films. In the other case the films were made on the fourth day and no spirochætæ could be found. He has included photographs of the two spirochætæ which he found in the set of photographs he has kindly sent us. We attempted to cultivate the spirochætæ directly from the blood-stream on several occasions from early cases, but with no success.

The urine.—Albuminuria was a constant occurrence in varying degree; some cases showed only a faint trace, and in others there was a considerable quantity. Casts, generally granular, were frequently found, and a few blood cells were almost always present in centrifuged deposits. Bile pigments in the jaundiced cases were present in large amounts; in some of the cases without jaundice bile could be detected

and in others it was absent. Acetone in considerable amounts was present in nearly all the severe cases during the pyrexial period. Leucin and tyrosin crystals were not found. We examined the stools of a series of cases for enteric group organisms but were unable to find them.

Morbid Anatomy.

Four cases terminated fatally which we were able to observe :—

Pte. M. Duration of disease unknown, died four days after admission with extreme jaundice and severe hæmorrhages. Skin showed multiple petechiæ and very pronounced jaundice of a bright yellow tint. Peritoneum, pericardium, and pleuræ covered with small bright-red hæmorrhages ; hæmorrhages in the heart muscle and under the endocardium. Heart soft and flabby. Hæmorrhages in the substance of the lung. Liver normal in size, colour, and consistency. Spleen small. Kidneys enlarged and swollen, with multiple hæmorrhages, and deeply bile-stained. Intestines and bile-passages normal. All the internal organs, where the colour permitted of its observation, were deeply tinged with bile. No spirochætæ found in liver by dark-ground illumination.

Pte. T. Died on twelfth day. Jaundice somewhat more intense than in the last case. Nothing abnormal noted with the exception of the large, swollen, congested, and hæmorrhagic kidneys. Intestines and bile-passages normal. No spirochætæ found in liver or kidney by dark-ground illumination.

Pte. H. Died on fourteenth day. Deep jaundice of skin and all viscera except the brain. Liver normal in size and colour. Spleen small. Kidneys large, swollen, congested, and showing multiple hæmorrhages. Stomach had two large polypi at the pyloric end, one of which had apparently been bleeding and looked like a large thrombosed pile. The heart was very soft and flabby. Two small hæmorrhages in the mesentery, one in a lymph gland. Duodenum and bile-passages normal. No spirochætæ found in emulsions of liver and kidney by dark-ground illumination.

Pte. E. Died on ninth day. Extreme jaundice of skin, no hæmorrhages. Lungs showed many diffuse, ill-defined hæmorrhagic areas, 1 to 2 cm. across ; they were most evident on the pleural surfaces, but were also well seen on the cut surface. One small hæmorrhage in the wall of the left ventricle. Heart soft and dilated with deeply bile-stained endocardium. Liver normal in size and colour. Spleen small. Kidneys swollen, enlarged, and showing multiple hæmorrhages. Intestine showed three patches of congestion of the mucosa, one of these in the duodenum. A few enlarged glands in the portal fissure and along the lesser curvature of the stomach. The bile-passages were free.

It will be seen that the hæmorrhages, in varying degree, occurred in three of the four cases. An interesting point is

that the brain was not bile-stained, though the cerebro-spinal fluid was bright yellow. The tissues from two fatal cases are available for study; two other fatal cases occurred, but in one instance the tissues were lost and in the other the tissues were sent home for microscopic examination, and we have no sections now in our possession.

The liver.—(1) Pte. E. (see Chart IX.); (2) Pte. H. (see Chart VIII.). Both livers showed a certain amount of diffuse granular degeneration, though hardly of significant extent. The most evident anatomical change, as seen in Case 1, is an exudation of polymorphonuclear leucocytes in the interstitial tissue surrounding the smaller bile-ducts. The leucocytes are diffusely scattered and nowhere suggest abscess formation, but around a section of small bile-duct 8 or 10 cells in length there may be seen scattered often as many as 12 or 15 leucocytes. They seem to be more numerous around the smallest interlobular ducts. Leucocytes may also be seen here and there between the liver columns, but nearly always within the blood-vessels. The liver (Case 1) shows a very considerable quantity of bile-pigment, mostly within the cells, in the form of greenish-black rods and granular masses. A difference in quantity between different parts of the lobule could not be definitely demonstrated. Bile-pigment was also found in the blood-vessels and in what we take, after some consideration, to be distended intralobular bile capillaries. These are most evident at the periphery of the lobule, in the neighbourhood of the smaller bile-ducts. Thus, a bile-duct may be seen at its origin from the lobule, surrounded by leucocytes, and just within the lobule at this point there may be seen, lying between adjacent liver cells, three or four spaces up to the size of a liver-cell nucleus, distended with bile-pigment, and apparently converging towards the bile-duct. Case 2 shows considerably less interlobular leucocytic exudate and considerably less bile. There is, however, a moderate amount of light-green pigment filling many cells in the form of fine granules. The structures which we take in Case 1 to be distended bile capillaries are much less evident here, although two or three small circular spaces have been seen, each lying between contiguous liver cells and containing a small amount of pigment. Leucocytes are seen in greater numbers than normal, but diffusely scattered between the liver columns and usually between the vessels. Thus, a row of four or five contiguous leucocytes in a small capillary is not difficult to find. Congestion in neither liver is pronounced.

The kidney.—The kidney in each case shows, apart from the acute lesions, a moderately extensive, chronic, diffuse cirrhotic process, rather more pronounced in Case 1. The acute lesions consist in, firstly, swelling and granular degeneration of the tubular epithelium, much more evident in the proximal convoluted tubules and the ascending limbs of Henle's loops. The distribution is diffuse, practically every tubule being involved, so that with most of the tubules the lumen appears to be obliterated. Secondly, a diffuse exudation of polymorphonuclear leucocytes in small numbers, lying between the tubules and more rarely within

them. Thirdly, hæmorrhage occurring in poorly defined patches usually involving several tubules. The blood is found mostly within the tubules and widely distending them, particularly in the distal convoluted portion. In both cases there was, however, considerable dilatation due to the chronic process, so it is difficult to decide in how far dilatation due to hæmorrhage occurred. All these changes were more marked in Case 1, where the degeneration of tubular epithelium was extensive. Glomeruli in neither case showed significant changes.

Sections of other organs showed no important changes, with the exception of the localised hæmorrhages in the lungs of Case 1 noted in the gross.

Spirochætæ in human tissue.—Sections of liver, kidney, adrenal gland, and lymph nodes have been prepared by Levaditi's older method and have been thoroughly searched for spirochætæ. We failed to find them in all tissues except the kidney of Case 1. In this kidney a small number of characteristic spirochætæ were found in isolated areas, so that in an entire section they would be found only in the neighbourhood of one glomerulus, but in considerable numbers at that point. They lie very occasionally between the tubules, but the characteristic place is the epithelium or lumen of a proximal convoluted tubule close to its origin from the glomerulus. Thus in one section of a tubule there may be found as many as 10. Where several occur together in this manner, the majority seem to lie near the centre of the tubule among the granular débris. The tubules in which they are found as a rule show more than the average amount of epithelial degeneration.

Experimental Results.

Tables I., II., and III. show the results of the intraperitoneal injection in the guinea-pig of blood of patients clinically diagnosed as Weil's disease. Table I. gives the results from cases showing definite jaundice; Table II. those from cases without evident external jaundice; and Table III. the combined results of the two series.

What we mean by a positive result is that the animal should show the typical signs of the disease, jaundice and hæmorrhages, and that we should find the spirochætæ in the blood or liver, either in films or in Levaditi preparations of the tissues. We have found, as the Japanese workers did, that the intraperitoneal injection of comparatively small quantities of blood gave the most satisfactory results. Various quantities have been tried, but our experience has led us now to employ as a routine a quantity of approximately 3 c.c. Larger quantities have not yielded better results and have often killed the animal on account of the overdose of foreign blood. We have injected as much as 7 c.c. without killing the animal, but there was severe shock and the result was negative.

TABLE I.—*Animals Infected from Cases of Weil's Disease which showed Definite Jaundice.*

Day of disease.	Number of attempts.	Average number of days for death of animal.	Positive.	Negative.	Percentage.
4th	5	10·5	5	—	100
5th	7	10·25	4	3	57
5th or 6th	2	8·5	2	—	100
6th	7	9·75	4	3	57
7th	4	11·5	2	2	50
8th	3	—	—	3	—
9th	2	—	—	2	—
—	30	—	17	13	56·6

TABLE II.—*Animals infected from Patients Clinically Diagnosed as Weil's Disease without Apparent Jaundice.*

Day of disease.	Number of attempts.	Average number of days for death of animal.	Positive.	Negative.	Percentage.
1st*	1	—	—	1	—
2nd	2	8	1	1	50
3rd	1	9	1	—	100
4th	4	9·5	4	—	100
5th	7	8·5	2	5	28·5
6th	4	22	1	3	25
9th	1	—	—	1	—
—	20	—	9	11	45

* No subsequent history of case.

TABLE III.—*Combined Results of the above Two Tables.*

Day of disease.	Number of attempts.	Average number of days for death of animal.	Positive.	Negative.	Percentage.
1st	1	—	—	1	—
2nd	2	8	1	1	50
3rd	1	9	1	0	100
4th	9	10	9	0	100
5th	14	9·37	6	8	42·8
5th or 6th	2	8·5	2	0	100
6th	11	15·8	5	6	45·4
7th	4	11·5	2	2	50
8th	3	—	—	3	—
9th	3	—	—	3	—
—	50	—	26	24	52

The Disease in the Guinea-pig.

The disease in the guinea-pig is characterised by pyrexia, jaundice, prostration in varying degree, and hæmorrhages in the skin, and in nearly all cases where the disease is allowed to take its course a fatal termination. It will be convenient to consider the findings under separate headings.

Incubation period.—The incubation period in guinea-pigs infected with blood from the patient varies within wide limits, and would appear to bear some relationship to the numbers and virulence of the spirochætæ in the blood, and possibly also to the presence of immune substances in the blood. The relationship, however, is not always easy to

follow, and apparently contradictory results are often obtained. Tables I., II., and III. show the average number of days from injection to death, in animals injected with blood taken on various days of the illness. It is necessary to note that the variation is almost entirely in the incubation period, the duration of the pyrexia being much more constant.

We have not included in the tables one guinea-pig which developed the disease 86 days after injection, although the animal was kept isolated throughout this period. This animal had been relegated to the stud as negative, and developed jaundice after becoming pregnant. Unfortunately, the identity of this animal was not established beyond all doubt, but from various corroborative evidence and the memory of the two men who cared for the animals there was every reason to believe that she was a guinea-pig which had been injected 86 days previously with human blood. The liver showed enormous numbers of spirochætæ, many more than usual. The fœtuses, about three-quarters term, were removed with the utmost precautions against contamination by maternal blood, and although no spirochætæ or lesions typical of the disease could be found in the fœtuses, an emulsion of the fœtal livers produced the typical disease when injected into a new guinea-pig. The amniotic fluid was clear and not bile-stained, and no spirochætæ could be found in the placenta, kidney, or lung (unfortunately the liver was lost) stained by Levaditi's method.

In passage animals the incubation period is much shorter and more constant, particularly after the third or fourth passage, and when very virulent strains are used the disease would seem to commence at times within a few hours of the injection of infected material.

The pyrexia.—The temperatures are taken morning and evening. As is well known, the temperature of a normal guinea-pig varies within wide limits, and is to some extent dependent on the external temperature. On this account it is often difficult to estimate the significance of a rise of temperature, but a little experience with guinea-pigs infected with this disease enables one to recognise, with few exceptions, the inception of the pyrexia which accompanies the infection. An important point is the comparison of the series of temperatures which are being recorded at the time. The temperature rises, sometimes with sharp spikes, often with only short steps, until the maximum is reached, and then falls sharply before death. The maximum temperature shows great individual variations, and one depends rather on the nature of the curve than on the absolute figure in recognising the pyrexia of infection. We insert four charts which are average examples. Two of these are animals infected from cases not showing jaundice and one is from a fatal jaundiced case. (Charts X.–XIII.)

In 24 guinea-pigs infected with human blood the average maximum temperature was 103° F., and in 53 animals infected with guinea-pig blood or liver (passage strains) it was 102·6° F. It may reach as high as 106° F. or more. The maximum once reached, the fall is very rapid, the downward curve on the chart being in the great majority of cases either a straight line or showing only one or two short steps.

CHART X.

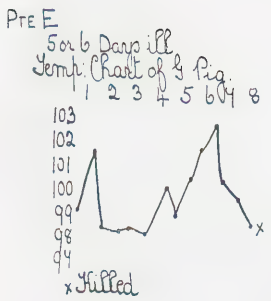


CHART XI.

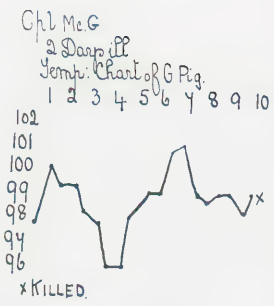


CHART XII.

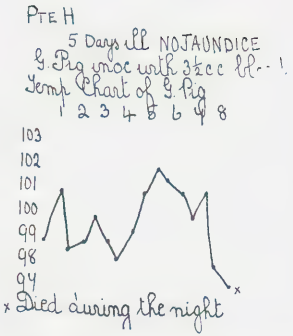


CHART XIII.

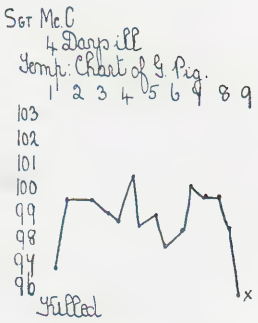


CHART X.—Animal infected from fatal case. (See Chart IX.) Spirochætæ demonstrated in Levaditi preparations of liver and kidney.

CHART XI.—Animal infected from non-jaundiced case. (See Chart VII.) Spirochætæ demonstrated in Levaditi preparations of liver and kidney.

CHART XII.—Animal infected from case with no jaundice. Spirochætæ demonstrated in sections of liver and kidney (Levaditi).

CHART XIII.—Animal infected from case with jaundice. (See Chart II.) Spirochætæ demonstrated in sections of liver and kidney (Levaditi).

The temperature becomes subnormal and the animal dies within a few hours. This is generally 12 to 36 hours after the maximum temperature has been reached.

The duration of pyrexia from the beginning of the rise to the collapse stage is remarkably constant. In 17 of 22 animals infected with human blood this time is between three and four days, and the average for the series is 3·5 days. In passage animals the duration of pyrexia is somewhat shorter, averaging 2·7 days in a series of 38 animals. It is to be noted that with the majority of our infected guinea-pigs we have not waited for death, but have killed them to ensure having fresh material for injection, since in a few cases failure has followed injection of material from guinea-pigs which had died during the night. The animals are not killed until they are moribund, with a subnormal temperature and in a state of extreme collapse. Many animals, however, have been allowed to die, and we have never seen an animal live more than six hours after the stage at which we call them " moribund " was reached.

Jaundice.—Jaundice does not appear, as a rule, until the temperature reaches its maximum; that is to say, only a short time before the collapse stage sets in. It is first obvious on the prepuce or labia, then in the sclerotics, and rapidly increases till the skin becomes deeply yellow. In the passage animals, when death supervenes rapidly, the jaundice is never so marked as when the pyrexia period is more prolonged.

Other symptoms.—The guinea-pig becomes ill when the temperature begins to rise, but the indisposition becomes more marked as soon as jaundice appears. The fur becomes ruffled, the animal refuses to eat and becomes thin and pinched, and as the temperature falls collapse sets in, with extreme prostration. At this stage the action of the heart is so weak that it is difficult to obtain blood from the ear veins for blood-counting purposes. Anæmia towards the end, when internal hæmorrhages become more extensive, is extreme. A small number of observations would seem to indicate that there is a definite initial leucocytosis. A subsequent progressive fall in the leucocyte count was observed, but the question as to whether this was not more than accounted for by the anæmia has yet to be settled by more thorough investigation of its relation to the red blood cells and hæmoglobin.

Pathological Changes in the Guinea-pig.

The post-mortem appearance in guinea-pigs which have died of the disease or which have been killed during the final collapse is very characteristic.

The skin is deeply and diffusely jaundiced, and jaundice is seen on all internal surfaces where the natural colour permits of its recognition. Numerous fine petechiæ are seen in the skin of the flanks and groin, and in the muscle layers of the abdominal wall and subperitoneally. The loose reticular and fatty tissues of the groins and axillæ show

more extensive and diffuse patches of hæmorrhage. Large hæmorrhages sometimes occur in the loose retroperitoneal fatty tissue, so that the kidneys, particularly the left, may be more or less completely covered by a thin hæmorrhagic layer. The small bowel usually, but not always, shows numerous superficial hæmorrhages, usually in the form of circular or elliptical patches, 2 to 5 mm. in greatest diameter, and situate opposite the mesenteric attachment. A single injected vessel is seen leading to the patch on each side of the bowel, and the patch itself is often outlined by a fine rim of injected, anastomosing vessels. In animals killed in the early stages of the disease, however, the bowel hæmorrhages are seen as minute pin-point spots situated at the side of the bowel at about the point where the encircling vessels commence to divide. These points evidently represent the commencement of the hæmorrhagic patch. Hæmorrhagic areas are also seen on the large bowel, and, in severe infections, on the stomach. In the latter situation they appear as groups of pin-point petechiæ. Enlarged, injected, and often hæmorrhagic lymph nodes are seen in the mesentery and in the groins.

The kidneys show very fine pin-point hæmorrhages under the capsule and through the cortex on section. The adrenal glands are often seen with such large hæmorrhages that all structure is obscure. The liver is more congested and of softer consistency than normal, but otherwise shows no gross changes. The spleen is congested, though not as a rule obviously enlarged. The lungs show the most characteristic gross change, being the seat of multiple, sharply defined hæmorrhages of varying size, which contrast sharply with the white lung tissue, giving the striking appearance which the Japanese authors aptly describe as being "like the wings of a mottled butterfly." The hæmorrhages vary from 1 mm. to 1 cm. in size, the larger ones being at the bases; when the hæmorrhages are numerous they may be seen throughout the lung tissue, but when they are few they are often nearly all subpleural. They may form a strikingly symmetrical double row along the lateral margin of the lower lobe, one row on each side of the sharp margin.

The hæmorrhages are an early occurrence in the infection, A guinea-pig killed exactly 24 hours after injection showed well developed lung hæmorrhages and beginning bowel hæmorrhages. It is to be noted, however, that the strain with which this animal was injected was at this time very virulent, and a control animal died on the third day. Four or five other guinea-pigs killed at stages from 36 to 72 hours after injection have all showed definite hæmorrhages.

Microscopic Pathology.

The liver.—The lesions seen in the guinea-pig's liver are essentially the same as those found in the two human livers—namely, uniform degeneration of liver parenchyma and an exudation of polymorphonuclear leucocytes, more extensive about the smaller bile-ducts. The degeneration varies greatly in degree, in some livers being very extensive, with considerable cell disintegration. As in the human liver, the

leucocytes are found both diffusely between the liver column and in localised collections about the bile-ducts, particularly about the smallest ducts at their origin from the bile capillaries. They are also found within the bile-ducts much more commonly than in the human liver. It is to be noted that polymorphonuclear leucocytes are more numerous than normal in the blood, as seen in the vessels of all tissues. No structures which could be interpreted as dilated bile capillaries have been found in the guinea-pig liver; the quantity of bile pigment is much less in the human livers, only occasional cells showing pigment, but these often being packed with it.

After considerable study of both human and guinea-pig livers we are of the opinion that the jaundice is to be ascribed to the inflammatory process noted about the smallest bile-ducts, which probably produces a partial obstruction. If this be so, the condition must in strict terminology be called an obstructive jaundice, the result of a cholangitis or pericholangitis.

Kidney.—The lesions here, as in the human kidney, give the picture of an acute exudative nephritis with localised hæmorrhages. The degeneration of the tubular epithelium may be very extensive, so that many tubules appear to be completely necrotic. The portions affected are, as in the human, the proximal convoluted tubules and the thicker portions of Henle's loops. Leucocytes occur in varying numbers. In severely affected kidneys they are numerous within and about affected tubules, and often immediately surrounding the glomerular capsule. Tubules are frequently seen distended with masses of leucocytes. Hæmorrhages are more discrete than in the human kidneys examined, often being limited, apparently, to individual tubules, the blood lying within the tubule, and in smaller quantity between the coils. A tubule may be seen distended throughout its entire visible length with free blood and thrombus. Tubules are also to be seen distended with homogeneous albuminous exudate. Glomeruli are not markedly affected, although occasionally one may find the tuft pressed back by a mass of thrombus, or still more rarely of leucocytes.

The lung.—Under the microscope there are seen, in addition to the hæmorrhages noted in the gross, numerous smaller ill-defined hæmorrhagic areas. Thus hæmorrhages involving only four or five alveoli may be seen. The larger hæmorrhages are often limited by the boundaries of the lobule, but may be larger or smaller. They are characteristically pyramidal in section, the base lying against the pleura, which may be raised over a considerable area by a layer of free blood. The vessels leading to the hæmorrhagic areas may sometimes be seen to be filled with leucocytes, or, again, a small mass of thrombus may be seen in the vessel, but the evidence is not sufficient definitely to indicate thrombosis as the cause of the hæmorrhage. A considerable amount of cellular proliferation may be seen more evident in the alveoli bordering on the hæmorrhagic

areas. Thus alveoli may be seen packed with large cells of endothelial character. Leucocytes may be seen in small numbers among these cells, or more rarely an alveolus is found filled with leucocytes. The bronchioles show no epithelial changes, but may contain leucocytes and débris.

The bowel.—Only one bowel hæmorrhage has been examined microscopically up to the present. In this case serial sections were cut through the entire hæmorrhage. These show the hæmorrhage to be associated with a large mass of lymphoid tissue, evidently, from its size and situation, a Peyer's patch. The hæmorrhage surrounds this completely in the form of a fairly thin capsule of free blood and lies entirely within the submucosa. A small amount of free blood is also seen throughout the lymphoid tissue. Through the exuded blood run widely distended capillaries, all apparently branches of the same arteriole and venule respectively—that is to say, in all the sections through the hæmorrhage only one main arteriole and venule are seen to pierce the bowel wall, and that approximately opposite the centre of the hæmorrhage.

Mesentery.—In one guinea-pig a small hæmorrhage, 1 mm. in diameter, was observed in the mesentery close to the small bowel. This portion of the mesentery was mounted entire, and the hæmorrhage was definitely seen to lie around the trunk of a small venule, the associated arteriole being quite free.

Spleen.—The spleen shows congestion and endothelial proliferation. The sinuses may be distended with large, clear, phagocytic cells containing red blood cells, leucocytes, or greenish-yellow pigment. Free pigment is also seen. These cells show not infrequent mitoses, and some show several irregular nuclei, or one giant nucleus irregularly lobed. Smaller spherical cells are also seen with a small, thin, crescentic nucleus at one border, the remainder of the cell being filled with packed red blood cells, or more commonly with eosinophile granules. These cells resemble strongly the "eosinophiles" seen in lymph nodes from cases of Hodgkin's disease.

Lymph nodes.—The lymph nodes show congestion and some endothelial proliferation, as seen in the spleen, but, save where hæmorrhages occur, no other significant change.

Adrenal gland.—In severe cases the adrenal is usually entirely hæmorrhagic, the hæmorrhage apparently originating in the medulla, and a certain amount of diffuse leucocytic exudation usually occurs.

Spirochætæ in the Guinea-pig Tissues.

The liver of the infected guinea-pig killed at the state of collapse usually shows very large numbers of spirochætæ. They lie mostly along the borders of the liver cells, more commonly at the junction of two cells, or several organisms may surround the cell like a garland, the description which the Japanese workers give. We find them, however, fre-

quently within the cells as well, although Inada and his collaborators say that this is rare, ascribing the fact to the absence of an immune substance in the blood of the guinea-pig. The spirochætæ, in some sections at least, are definitely more numerous at the periphery of the lobule and in the neighbourhood of the smaller bile-ducts. They may be also seen in small numbers in the connective tissue around these small bile-ducts, and not infrequently within the epithelial cells or free in the lumen of the smallest ducts.

In the kidney the distribution of the spirochætæ is characteristically in discrete areas, as noted in the human kidney, so that individual tubules or groups of two or three may show large numbers, the intervening tissue being free. The site is usually within the epithelial cells or free within the lumen of a proximal convoluted tubule close to its glomerulus. Often a mass of many spirochætæ may be seen lying among the granular débris at the centre of a degenerated tubule, and the impression gained is that multiplication has taken place in this situation. Spirochætæ may also be seen occasionally in the intertubular tissue. We have never seen a spirochæta in glomerular tuft or lumen.

We have found spirochætæ in the adrenal gland, lying within the epithelial cells of the capsule. We have also found them in the spleen (fairly numerous), lymph nodes, and in the heart muscle. We have never succeeded in demonstrating them in the lung.

In the blood of the guinea-pig they are fairly numerous, especially in the passage strains where one can find them in films in every third or fourth oil-immersion field. We have never seen them so numerous in the blood as would be expected from the description given by the Japanese authors, and we have not been able to find them with dark-ground illumination in the blood unless it was first hæmolysed with distilled water or saponin and then centrifuged.

The site of multiplication of the spirochætæ has not been determined, but from the numbers found in the liver it may be that it is in that organ that they grow. If multiplication takes place in the blood stream, a possible explanation of the numbers found in the liver, and around the proximal convoluted tubules in the kidney, is the relatively slower circulation at those points.

We have found that the older Levaditi process is the more reliable method of demonstrating the organisms in the tissues; on two occasions pieces of tissue prepared by the pyridin method did not show the organisms, while with the older method they were easily found. For films from the blood or liver we have found Fontana's method the most reliable and rapid, while with Giemsa the time taken for satisfactory staining is often 36 hours, and the organisms are not so distinctive or so easily found. With dark-ground illumination we have been disappointed, although the apparatus we use—a Leitz stage and a small arc lamp—shows *S. pallida* very well. With dark-ground illumination they are much less refractile than *S. pallida* or *S. refringens*. Their movements are slow and undulating, and the most

characteristic movement is from side to side at one or both ends, a central portion remaining rigid. A slow spiral movement is occasionally seen. The spirochætæ show pronounced cross striation, caused, apparently, by a uniform series of refractile "beads" and suggesting the appearance seen in striated muscle.

The spirochæta as seen in the living condition or stained is of inconstant shape and has no fixed undulations. The thickness is about that of *S. pallida* or slightly less, and the length varies considerably, being on the average about the length of a specimen of *S. pallida* showing 12 to 14 turns. In films from liver or blood the spirochæta is often nearly straight or shows only wide undulations, while the ends in the majority of organisms are seen recurved like a fish-hook. There is a definite taper to each end from the central thicker portion. In the tissues prepared by Levaditi's method the spirochætæ often show angular, irregular undulations, and the tapering at the ends is not so well seen.

We are greatly indebted to Dr. A. C. Coles (Bournemouth) for the photomicrographs which he has kindly taken for us and given us permission to use. They are taken by his special method and illustrate the appearance of the spirochæta in films very well. He has included specimens of *S. pallida* and *S. refringens* for comparison. The photographs of the organisms in the tissues have been done for us by Dr. J. T. Wigham (Trinity College, Dublin) and are also characteristic. We owe our best thanks to these helpers.

Technique of Passage.

In the passage of the disease from animal to animal the blood taken from the heart or an emulsion of ground-up liver is generally successful. We have found, contrary to what Inada states, that the liver emulsion is more reliable than the heart's blood. On three occasions the liver has passed the infection where the blood failed.

The liver is crushed in a mortar and to about 10 gm. of liver is added 5 c.c. of saline, and the whole is briefly centrifuged to throw down particles which could block the needle. The supernatant fluid is then injected intraperitoneally, using about 2 c.c. for each animal.

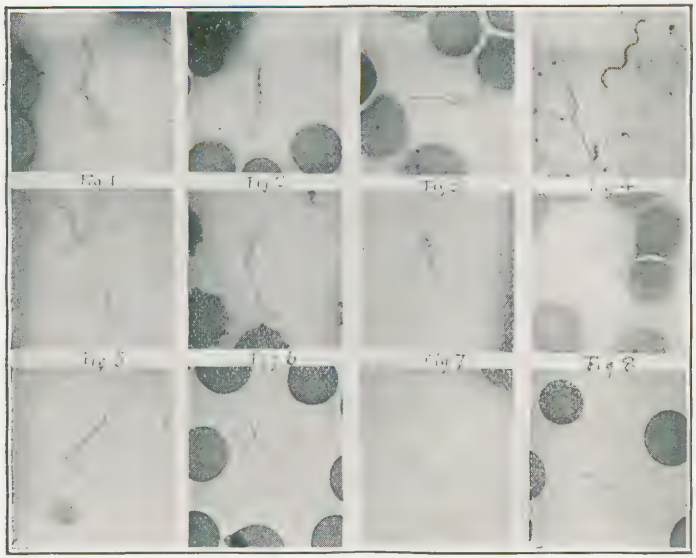
The injection should be intraperitoneal to be satisfactory, since subcutaneous inoculations give a longer incubation period, and in two or three cases we have seen abscesses or spreading infection of the subcutaneous tissues at the site of subcutaneous injection. Passage has also been performed successfully by spreading liver emulsions on the shaved skin and by feeding the animal on carrot steeped in liver emulsions. We have also passed the infection with the urine of the killed animal. One attempt to infect with the bile failed.

Virulence.

The virulence of the spirochætæ is very strikingly increased by passage.

The average period from injection to death in 23 animals infected from human blood was 10 days. The average time for death in 14 passages of one strain was 4·8 days, and of

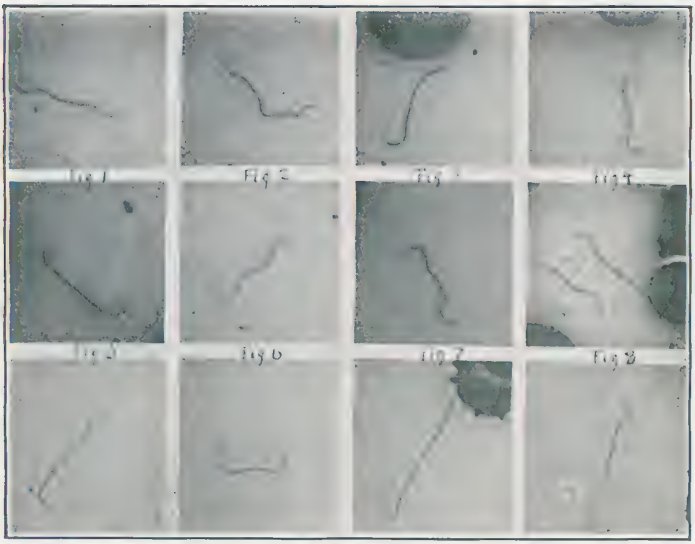
PLATE I.



Shows 11 examples of *S. ictero hæmorrhagica* in films of blood from infected guinea-pig. Multiplied 1000 diameters. Stained by Giemsa's stain. 1 example of *S. pallida* and *S. refringens* for comparison. Photomicrographs (I., II., and III.) by Dr. A. C. Coles, Bournemouth.

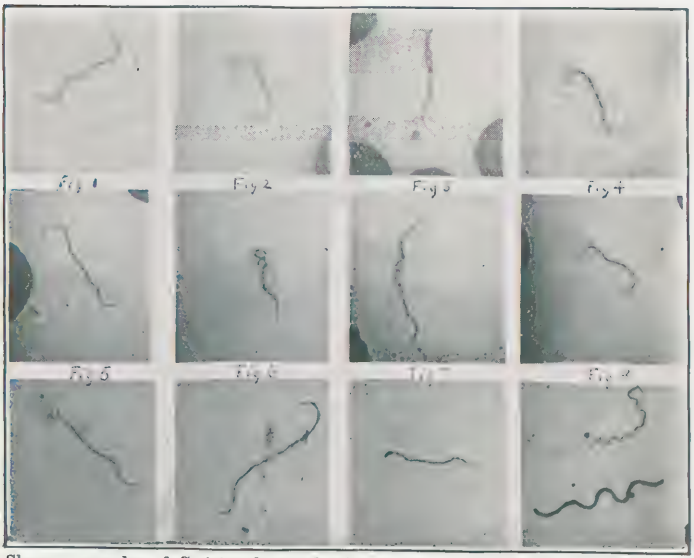
another strain, for 11 passages, was 5·3 days. This period for the first two or three passages may vary somewhat, but is usually 6 to 8 days; in succeeding passages it becomes progressively shorter and more constant, until a point is reached after 5 or 6 passages at which it becomes approximately fixed at 4 days. With small animals, when the strain is virulent, we have had them die on the third day, but this has always been in half-grown animals. In these young guinea-pigs the infection seemed to be overwhelming. The temperature reaction was slight and the jaundice not well

Plate II.



S. ictero-hæmorrhagica. × 1500. Various forms from blood of guinea-pig

Plate III.



Shows examples of *S. ictero-hæmorrhagica* in blood films. Nos. 2 and 3 on this plate are *S. ictero-hæmorrhagica* from human blood, Corp. MacG. (see Charts VII. and II.). Magnification 1500 diameters. Stained by Geisma's stain.

developed. The duration of the pyrexia, as already mentioned, was definitely shorter in passage animals, and this becomes more marked in the later passages.

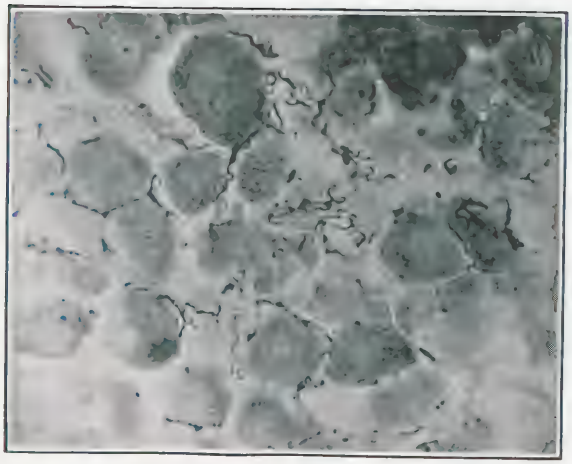
Jaundice does not have time to develop to the same extent as where the disease is of longer duration. Otherwise the post-mortem appearances show no important difference. The hæmorrhages are often more extensive but this is not constant. Another criterion by which the increase in virulence may be judged is the number of spirochætæ found in films from the blood and liver. In guinea-pigs dying from infection with human blood one may fail to find them in the blood films after 30 minutes' search of specimens stained by Fontana's method, while the liver films may show them only in 8 or 9 fields. In some animals infected from man we have only succeeded in finding the spirochætæ in sections of tissues with Levaditi impregnation. In the animals of the latter passages it is always easy to find the spirochætæ in the blood, although they are seldom numerous, but the livers of these animals show large numbers, 10 to 12 spirochætæ in every oil-immersion field of a film made from the cut surface of the liver.

Experiments with Urine.

The Japanese authors showed that the spirochætæ, both in the experimental disease and the disease in man, were excreted in the urine, the excretion commencing soon after the blood began to develop a demonstrable immunity and continuing in certain cases up to the fortieth day from the onset of illness. They were also able to infect animals with the urine of patients, 3 of 9 inoculations being positive when the injection was done before the tenth day, and 5 of 15 positive when the urine was taken between the eleventh and twenty-first days. They were able to find the spirochætæ in the urine with dark-ground illumination and regarded the excretion in the urine as a most dangerous source of infection. We have so far failed to find spirochætæ in the urine and to infect animals with urine, although we have repeatedly searched for them and tried various methods of demonstrating them. It must be remembered that the patients were for the most part evacuated at the end of the pyrexial period, tenth to fifteenth day, and it is after this time that the spirochætæ become abundant. We are informed that the spirochætæ have been found with comparative ease at the base hospitals at a later period of the disease. In spite of this we think that our failure to get positive results at the earlier stage of the disease is of some significance, and think it is possibly due to the comparatively benign type of Weil's disease prevalent in this country. We have only in a few cases been able to examine the urine as late as the fifteenth day. We append a list of the experiments.

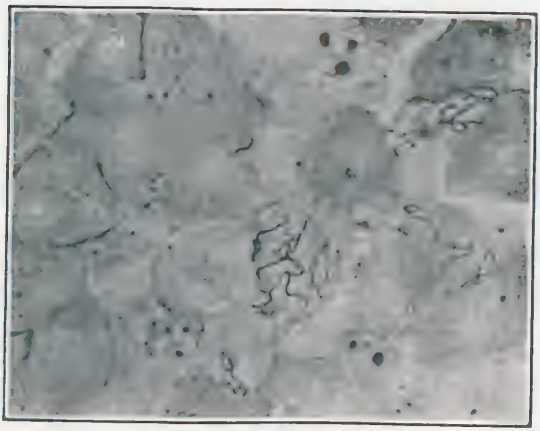
The animals were inoculated intraperitoneally with the centrifuged deposit from the amount of urine mentioned.

FIG. 4.



Liver of guinea-pig showing *S. ictero-hæmorrhagica*. Sections prepared by Levaditi's first method. Dr. J. T. Wigham.

FIG. 5.



Liver of guinea-pig showing *S. ictero-hæmorrhagica* (Levaditi). Dr. P. P. Laidlaw.

They were inoculated with the urine of the same patient on two successive days, and were watched and had their charts kept for 14 days.

G.-p. 21 ...	...	150	c.c. on	14th	and	15th	day of illness	(Pte. G.)
,, 22 ...	...	300	,,	10th	,,	11th	,, ,,	(Pte. H.)
,, 23 ...	...	150	,,	13th	,,	14th	,, ,,	(Pte. C.)
,, 24 ...	...	150	,,	15th	,,	16th	,, ,,	(Pte. MacD.)
,, 43 ...	...	170	,,	13th day of illness				(Pte. L.)
,, 52 ...	...	320	,,	13th	and	14th	day of illness	(Pte. B.)
,, 64 ...	...	320	,,	7th	,,	8th	,, ,,	(Pte. C.)
,, 67 ...	...	300	,,	7th	,,	8th	,, ,,	(Pte. T.)
,, 74 ...	...	300	,,	6th	,,	7th	,, ,,	(Gnr. W.)
,, 87 ...	...	200	,,	11th day of illness				(Pte. B.)

The results were consistently negative. In one of the fatal cases there was suppression of urine for 24 hours before death, and we were unable to secure a specimen to inoculate an animal. In the second fatal case which we were able to investigate there were not sufficient animals available at the time to try to infect with the urine.

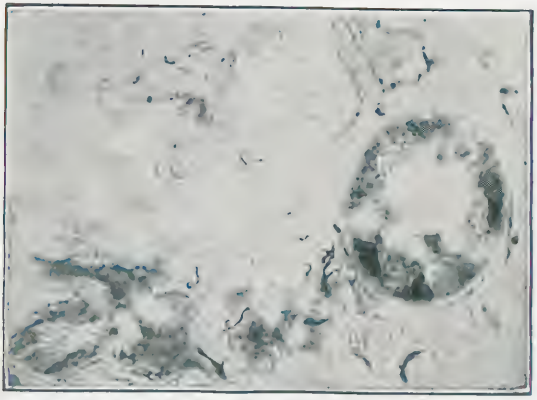
Immunity.

In their first paper on infective jaundice the Japanese authors showed that the blood of convalescent patients contained substances capable of curing infected animals and of protecting them against infection ; they dated the appearance of this immune substance at about the tenth day of illness, at which time it first became demonstrable. In the later paper, the proofs of which we have been allowed to see through the kindness of Dr. Flexner, director of the Rockefeller Institute, they report that they have succeeded in immunising a horse and in using the serum therapeutically with encouraging results. The following experiments were done in confirmation of the original Japanese paper :—

Experiment 1.—Guinea-pig 35. The animal developed jaundice eight days after injection, the temperature being on the downward grade after reaching 103·2° F. The animal was then injected with 2½ c.c. of serum from a patient on the fourteenth day of illness. It showed immediate improvement and began to feed, and the fur became normal ; all jaundice was gone on the eleventh day. On the fourteenth day 2 c.c. of liver emulsion rich in spirochætæ was administered and did not infect the animal. This injection was repeated on two subsequent occasions with virulent strains and the animal remained immune.

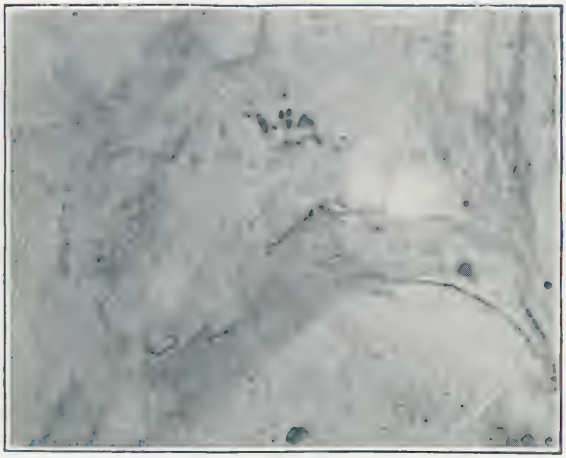
Experiment 2.—Guinea-pig 40. The animal was injected at the same time with 2½ c.c. of serum from a convalescent patient, and 1 c.c. of liver emulsion with 1 c.c. of heart's blood from an animal which had died of jaundice. No symptoms developed for 14 days. A control animal died in 7 days. Guinea-pig 40, reinoculated on the fifteenth day, died four days later with jaundice and typical signs. This would indicate that passive immunity is transient, and that,

FIG. 6.



Kidney of guinea-pig showing *S. ictero-hæmorrhagica* (Levaditi). Dr. P. P. Laidlaw.

FIG. 7.



Kidney showing *S. ictero-hæmorrhagica* in human tissue. (Pte. E. See Charts IX. and X.). Dr. J. T. Wigham.

although the animal was protected by the immune serum, it did not develop any active immunity, since infection had not occurred.

Experiment 3.—Guinea-pigs 41 and 47 were subjected to a repetition of Experiment 1, and the result was identical.

Experiment 4.—Guinea-pigs 45 and 46 were treated with immune and normal serum respectively when the collapse stage had been reached. Both died within three hours.

Experiment 5.—Guinea-pigs 83, 85, 86, 89, and 90 were each injected with the same quantity of virulent liver emulsion, and at the same time human serum was administered as follows:—

Guinea-pig 85.—3½ c.c. normal human serum.

Guinea-pig 83.—2½ c.c. serum from jaundice case taken on sixth day of illness.

Guinea-pig 89.—2½ c.c. of serum from same case taken on eighth day.

Guinea-pig 86.—2½ c.c. of serum from same case taken on tenth day.

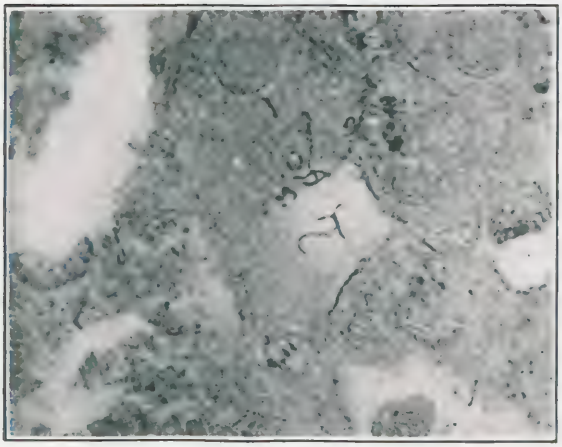
Guinea-pig 90.—2½ c.c. of serum taken from same case on twelfth day.

Guinea-pigs 85 and 83 died on the fifth day with typical signs of the disease. Guinea-pig 89 died immediately after injection. Guinea-pigs 86 and 90 developed no symptoms.

In only one respect do the results of these experiments differ from those obtained by Inada and his assistants, who state that the serum must be given before the appearance of the jaundice in order to cure the animal. We have found that the serum of a convalescent patient will cure the animal even after the appearance of jaundice, if it be given before the collapse stage is reached.

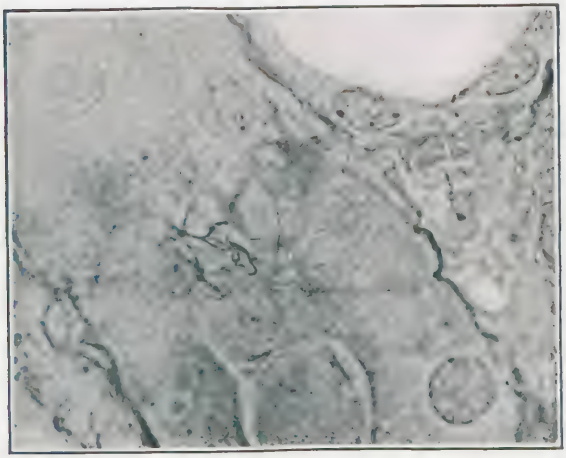
After these experiments had been done we suggested the immunisation of a horse with a view to treatment, but the suggestion was not put into practice, partly owing to the fact that the prevalent type of the disease was relatively benign and did not seem to call for very active treatment. The Japanese workers, in their treatment with serum, succeeded in considerably lowering the death-rate, which was high (32 per cent.), but did not shorten the duration of jaundice. They also were apparently able to rid the circulating blood of spirochætæ and to hasten the appearance of the immune reaction in the blood. They found that rather large doses of the horse serum were necessary, 60 c.c. or more, and that better results were attained by early administration. This would indicate that a very high titre serum would give better results. They found that the serum which they had prepared was about as potent as that of a convalescent case. They have further attempted to produce

FIG. 8.



Liver of guinea-pig showing *S. ictero-hæmorrhagica*. Animal infected from case that did not develop jaundice. Dr. J. T. Wigham.

FIG. 9.



Kidney from same animal as Fig. 8. Dr. J. T. Wigham.

active immunity by the injection of carbolised cultures, but the results are not as yet convincing.

Sero-diagnosis.

As soon as it became clear that there were considerable numbers of cases of spirochætosis which did not show obvious jaundice, it seemed to us desirable to find some convenient form of laboratory diagnosis other than the infection of animals. With this end in view a large number of experiments were done with a view of finding whether this could be attained by a complement-deviation test. Antigens were prepared in various ways from the livers of guinea-pigs with very great numbers of spirochætæ, but no test sufficiently reliable could be elaborated. There was nearly always a difference in the hæmolysis which held out hopes of arriving at a reliable test, but there was not sufficient constancy. For one instance immune guinea-pigs did not give a positive reaction as compared with normal animals. A further point was that the bile, both in the serum and in the antigen, produced strong anti-complementary action.

Cultures.

Inada and Ito succeeded in cultivating the spirochætæ in 1915, and state that they got their original cultures by Noguchi's method.

Ito and his collaborators publish a fuller account of the cultivation of the organism in the *Journal of Experimental Medicine* for April, 1916, and have had no difficulty in cultivating the spirochætæ by taking the heart's blood of an infected animal and growing the blood in deep blood agar or blood gelatin tubes. They state that the cultures begin to grow in 8 days, the maximum being reached in 16 to 20 days. So far as we are concerned, we have been unable to obtain cultures, although we have attempted it repeatedly in different ways and at different temperatures. We tried the original Noguchi technique with guinea-pig liver and kidney as the tissue component, with blood, ascitic fluid, or guinea-pig blood as the enriching medium, and with agar or broth as the basis; in all cases the results were negative. We have tried different infecting agents, blood, liver, or kidney from animals with jaundice, and never succeeded in getting an undoubted culture. Though we have often thought that we could demonstrate the spirochætæ by staining with Fontana's method, we could not see them in such cultures with the dark-ground stage, while, conversely, on one occasion we thought that we had them in culture after examining by the dark-ground stage, but were unable to stain the organism. We attempted several times to cultivate direct from the

patient, taking early cases and a relatively large amount of blood into deep agar tubes and covering with paraffin, but these efforts were also abortive.

We mention these negative results as showing that cultivation is not so simple a task as might be supposed from the literature. The possibility of an inherent difference in the organisms is to be considered.

Experiments with Lice.

When we began to consider the route of infection in the soldier our first idea was the possibility of lice being the infecting agent, as they may be said to be invariably present on soldiers serving in the field. Some colour was lent to this idea by the fact that we had never seen an officer with true infectious jaundice. The Japanese authors had considered the possibility of fleas and mosquitoes being the intermediate host and discarded both. The fact that the majority of cases came from a relatively small area, that jaundice was rare except in one part of the line, as will be more fully explained later, and that divisions which left the infected area did not have any more cases of jaundice, suggested that lice were not responsible. On the other hand, the analogy of relapsing fever made it necessary to attempt some experiments on the matter. The lice seldom survived three days' separation from the human host and seemed not to thrive on guinea-pig blood.

Guinea-pig 88.—40 lice were fed on an animal which had spirochætæ in the blood-stream, demonstrated by films; on the next day the survivors were fed on a fresh animal (Guinea-pig 88), and again on the following day the survivors were fed on this same animal. Result negative.

Guinea-pigs 97 and 98.—The same as above.

Guinea-pig 122.—50 lice were allowed to feed on a patient on the fifth day of his illness, and were then fed alternately on the patient and the animal till there were no survivors at the end of 60 hours. Result negative.

Guinea-pig 124.—The same experiment with another patient on the fourth day of illness.

Guinea-pig 123.—100 lice were taken from the clothing of a patient on the sixth day of illness, crushed and emulsified in saline, and injected into the peritoneal cavity of the animal. Result negative.

Guinea-pig 129.—Same experiment as the last with 40 lice. Result negative.

Guinea-pig 125.—Lice taken from the clothing of a patient on the ninth day of illness were fed on the animal till there were no survivors. Result negative.

There is an obvious fallacy in these experiments which is hard to eliminate. The incubation period in the louse may be prolonged and the parasite not become infective for several days, the lice dying, when kept in captivity, before the infective period is reached. Further, the lice taken from a man may not have fed on him while the spirochætæ were abundant in the blood-stream. The experiments, however, so far as they go, bear out what would have been expected from the almost strict area from which the cases came. A large number of lice have also been examined by Sergeant Peacock, R.A.M.C., by staining methods with a view of finding the spirochætæ. The lice were either taken from men with jaundice or deliberately infected from animals with spirochætæ demonstrable in their blood, and all the examinations were negative.

From every point of view we think that it is improbable that lice are the intermediate hosts if there be an intermediate host in the cycle of infection. We have tried no experiments with mosquitoes, and though the incidence went up as the warm weather set in and has declined with the colder weather, when mosquitoes are scarce, it is not sufficient evidence to incriminate these insects. There were, however, undoubted cases during last winter. The restriction within narrow limits of the epidemic is also in opposition to the hypothesis of transmission by the mosquito.

Epidemiology.

There were, as we are informed, undoubted cases of Weil's disease during the winter of 1915–1916 which were seen at the base hospitals, but the diagnosis was not confirmed by animal inoculation. So far as we know, the first cases which were recognised in this area occurred at the end of April and the beginning of May, 1916. The number of cases varied from time to time. There were more cases seen during the early part of July and the end of August than at any other periods, though there were a few cases passing through during the whole summer. At one time there were 15 cases under observation, and others had been evacuated on admission as sufficiently well to travel. The incidence seemed to vary with the weather conditions. Thus during and immediately after a dry, warm spell of summer weather there were very few cases, but with the advent of rainy weather the incidence went up and many cases were admitted. On two occasions this occurred, the most striking being after the dry spell which prevailed during the end of July and the first fortnight of August. From August 1st to the 23rd there were no cases of jaundice, and from August 23rd to Sept. 1st 12 cases were admitted. Rain fell in considerable quantities from August 13th to the end of the month.

We know of only one officer who definitely developed Weil's disease, all the cases having occurred among the other ranks and practically always among the "trench troops." The men always either developed the disease in the front line or very soon after they had been relieved. Certain divisions in a corps were affected, others being free. Within the divisions certain brigades were affected, and within the brigades certain regiments had more cases than others. We found that a regiment which had a number of cases while in the line was not infected while in rest billets, but again produced cases when it returned to the trenches. There were certain areas from which cases came, and if we represent the line from which we collect as a straight line divided into six segments, numbered from left to right, Segment 1 and Segment 4 provided the great majority of cases.

By the kindness of Colonel Gerrard, A.M.S., we were allowed to see a "spot map" of Sector 1, which had been kept for about two months, the map representing the cases as they occurred among the troops in occupation of the area, and giving the place where the affected men had been on duty. This map showed that the cases in the infected area were grouped round two small sections of trench, one at each end of Sector 1.

A new division, which had, up to the time of taking over the infected area, never had a case of jaundice, developed cases of jaundice within 14 days of going into the new trenches, and the same division, after being relieved and going to a new part of the line, was again free from cases. Again a new division took over the infected area, and in the same way cases of jaundice cropped up within a short time.

The trenches in this infected area were wet, not well drained owing to the nature of the ground, and in some places, even in dry weather, never properly dried. This was so in both the segments of the line from which cases came. This is suggestive when one remembers that Inada and his collaborators state that cases of jaundice occurred in the wet shafts of a mine, and that the men working on the surface and in the dry shafts were not infected, and, further, that when these wet shafts were pumped out on their advice the incidence markedly decreased. They also suggested that possibly the entry of the organisms might be through the skin—a guinea-pig can be infected through the skin—but they were of the opinion, after a general review, that the mode of entry was through the alimentary canal. A point

of interest is, though it is complicated by the contemporary onset of colder weather, that the incidence of the disease in our area had practically ceased, and that the troops were issued with high "gum" boots a week before we saw the last case of jaundice. Since writing the last sentence two cases have been noted.

In the proofs of a forthcoming paper by the Japanese workers which was sent to us by Dr. Flexner it is stated that they have succeeded in finding the spirochætæ of jaundice in the kidneys and urine of 38 per cent. of ordinary field-rats in the infected areas in Japan. They suggest that the infection may be conveyed by the rats' urine, either directly or indirectly. We have not yet had time to confirm these findings, but hope soon to be able to do so.

Cases have occurred in the Allied troops in different parts of the front. Dr. L. Martin has published an account of some cases investigated by him in the French Army, and we also know of other cases which have occurred among the troops in Flanders which have also been proved experimentally. Whatever be the portal of entry of the spirochæta, it would appear that wet and muddy trenches are a predisposing factor. If the infection had been water-borne there would have been a more widespread epidemic. If vermin or mosquitoes had been the infecting agents the epidemic could hardly have been so localised, and the experiments which we have performed, such as they are, would seem to exonerate them. We would also add that there have been up to now no ward or laboratory infections. The patients have been cared for in a general ward with no further precautions than would be carried out if they were enteric cases.

Conclusion.

The clinical and experimental examination of about 100 cases of Weil's disease which we have been in a position to investigate during the last six months justifies, we think, the conclusion that the disease, as it has been observed among British troops in Flanders, is identical with that described by Inada and his collaborators in Japan.

The virulence of the infection in the cases seen here is much less than that described in Japan, which is well shown by the comparatively low mortality. All the essential findings of the Japanese authors have been confirmed, save that our cultural attempts have so far failed.

In addition we have noted the occurrence of cases of spirochætosis which have not shown any external jaundice.

The success of our work is to a great extent due to the efficiency and zeal of our laboratory attendant, Sergeant B. Baker, R.A.M.C. (T.F.).

After-note.—Since writing the paper we have been able to confirm the statement of the Japanese workers mentioned in the paper with regard to the finding of the spirochætæ in the kidneys of field-rats. Of 9 rats taken from the right part of Segment 1 in the chart in the paper, 5 proved infective to guinea-pigs. Of 6 rats from the left end of Segment 1 one communicated the disease. The kidney of the rat was crushed and emulsified, and injected intra-peritoneally into the guinea-pig. The disease in the infected animal was typical, and we have found the typical *S. ictero-hæmorrhagica* in the organs of the guinea-pig. Levaditi preparations of the rat kidney have shown the spirochætæ, and we have also found them in films made from an emulsion of the kidney stained by Fontana's stain.

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